Estrogens still represent an attractive therapeutic approach for Alzheimer’s disease

Alzheimer’s disease (AD) is a progressive neurodegenerative condition that goes from mild cognitive impairment in prodromal disease to severely disabling deficits in advanced stages. The risk for AD development, as well as progression and severity, clearly differ between men and women (Pike, 2017). Epidemiological studies have shown that there is a significantly increased prevalence in the development of AD in women compared to men, which is usually explained by the longer lifespan of women. This increased frequency may be due to the interplay between age and sex, in which genetic factors together with hormonal and metabolic patterns play a crucial role. Moreover, cognitive impairment has been confirmed to be greater in women than in men at the same stage of AD, likely due to reduced estrogen levels in post-menopausal women (Laws et al., 2016).

From the beginning of the 1990s, when the first evidence of a beneficial role of sex hormones on cognition appeared, to today, the interest of estrogens as neuroprotective agents for AD has had highs and lows due to a large disparity of data in the literature. Thus, excellent results have been obtained from basic and epidemiological studies but these have not been confirmed by clinical trials. In fact, the first clinical trials, in contrast to what was expected, have seen an increased risk of dementia in post-menopausal women who have undergone hormone replacement therapy (Shumaker et al., 2003).

In AD, estrogen neuroprotection seems to be exerted at multiple levels. Despite their classical protective action against neuroinflammation, synaptotoxicity and oxidative stress, recent findings demonstrate that estrogens are able to modulate the production of the two protagonists of the disease: amyloid-β (Aβ) and Tau protein.

Aβ derives from the amyloid precursor protein (APP) through β site APP cleaving enzyme 1 and γ-secretase processing that target Tau protein has been extensively studied. Tau is a microtubule-associated protein characterized by multiple highly regulated phosphorylation sites. The dysregulation of Tau phosphorylation leads to accumulation of its hyperphosphorylated form, which aggregates and forms intracellular deposits, named neurofibrillary tangles. It has been demonstrated that 17β estradiol promotes Tau dephosphorylation in vitro in rat cortical neurons and neuronal cells in an estrogen receptor-mediated and dose-dependent manner. Also in vivo studies have shown that estrogenic treatment activates signal pathways that lead to an inhibition of kinases such as GSK3beta and therefore to a reduction in Tau-phosphorylation (Munoz-Mayorga et al., 2018).

Since Aβ and Tau could be targeted by estrogens at different levels, the most recent literature in the field has dedicated attention to the relationship to Aβ/Tau interactions, and thus, in vivo, the two proteins are reciprocally involved in pathological signals. Several data support the amyloid hypothesis: accumulation of Aβ peptides is the primary and early event that induces neuronal degeneration, characterized by altered and aggregated Tau.

We have developed a powerful system based on mice expressing the wild-type human Tau (hTau) which were subjected to intraventricular injections of Aβ peptides, in nanomolar concentration. We discovered that Aβ42 monomers, but not oligomers are able to produce PHF-like conformation of Tau protein, and to induce two phosphorylated epitopes which are not present in normal Tau (Ser396 and Ser422) through the activation of GSK3β, JNK and ERK 1/2 kinases in male hTau mice (Manassero et al., 2016).

Aβ42 induces phosphorylation of the pathological sites in male and ovariectomized female mice (Ser396 and Ser422) followed by complete protection of Tau phosphorylation (Manassero et al., 2016). Moreover, the two proteins are not able to determine this effect in young female mice but also after ovariectomy. The same result was obtained by evaluating the total Tau protein levels.

We also showed that the treatment with Aβ42 induces phosphorylation of the pathological sites in male and ovariectomized female mice, while controlled female’s phosphorylation of the sites is not observed. To confirm whether the presence of estrogens is involved in the different effect exerted by the treatment with Aβ42 on the pathological conformational change of Tau, groups of female mice, ovariectomized or not, were subcutaneously treated with estradiol (1 μg/kg) and fed with a phytoestrogens free diet for 3 weeks. As expected, oophorectomy significantly decreases circulating estradiol levels, whereas the treatment with estradiol completely protects both the pathological conformational change and the increase of total Tau mediated by Aβ42 in ovariectomized females.

The enrichment with estradiol is also followed by complete protection of Aβ42-mediated phosphorylation, after oophorectomy, of pathology-related sites. Finally, to further confirm the role of estradiol on the pathological conformation change and hyperphosphorylation of Tau, we also treated male mice with estradiol and found that this treatment is able to completely protect both the conformational change and the hyperphosphorylation of Tau (Guglielmotto et al., 2020).

Literature data indicate that estradiol treatment, at least during the early stage of AD pathology, significantly promotes the recovery of cognitive function and upregulated neurogenesis-related mediators in Aβ42 mice and that these effects may have been due, at least in part, to decreased levels of oxidative stress (Nilsen, 2008). Thus, we tested the total antioxidant capacity and found that ovariectomy is capable of causing a significant decrease in antioxidant capacity and the simultaneous intracerebroventricular injection of Aβ42 induces a further deterioration of the parameter. Treatment with estradiol protects the drop in antioxidant capacity by bringing it back to control values, confirming an antioxidant role of estradiol in our experimental model (Guglielmotto et al., 2020).

Finally, we measured levels of miR-218, since recent discoveries demonstrate that estrogen receptors are able to modulate the expression of microRNA involved in Tau phosphorylation (Xiong et al., 2015). In particular, it has been found that an increase of miR-218 reduces the level of target protein tyrosine phosphatase a with consequent enhancement of Tau phosphorylation.

We observed that levels of miR-218 are significantly higher in ovariectomized female mice, injected or not with Aβ42, whereas the estradiol treatment is followed by a total protection of the miRNA increase.

The fact that the regulation of miRNAs plays a role in many pathological conditions of the central nervous system may open new windows for the research on the role of estrogens in AD.

Biological complicity of miRNA is only shortly known but it is now quite evident that these short RNAs have an important role in modulating and regulating gene expression. In the literature, there is a lot of data regarding the role of estrogens in the regulation of miRNA in cancer studies while the role of estrogen regulation in the brain is still very unexplored. A first study of this field by Rao and collaborators (2013) showed that estradiol is able to regulate target miRNA in age and tissue specific way in ovariectomized rats. Furthermore, prolonged estrogen deprivation leads to a loss of
Inhibition of amyloid-β (Aβ) mediated pathological conformation of Tau by estradiol treatment.

Estradiol replacement protects against the pathological conformation of Tau mediated by monomers of Aβ. This process involves both antioxidative activity as well as by its ability to modulate the expression of miRNA-218 linked to Tau phosphorylation.

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**Perspective**

**Aβ\(_{1-42}\) monomers**

**17 β ESTRADIOL**

**Protection of oxidative stress**

**miRNA-218 modulation**

**Tau hyperphosphorylation**

**Tau aggregation**

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**Neurodegeneration**

![Figure 1](https://doi.org/10.4103/1673-5374.314295)

**Figure 1** | Inhibition of amyloid-β (Aβ) mediated pathological conformation of Tau by estradiol treatment.

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**References**


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